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The effect of ecological forest restoration on bumblebees (*Bombus* spp.) in the boreal forest

*Effekter av ekologisk restaurering på humlor (*Bombus* spp.) i
boreal skog*

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Abstract

*Insect pollinators are an important part of biodiversity, yet the number of pollinating insects is declining all over the world. Of all the wild plant species about 80% are reliant on insect pollinators, with bees being most important. One of the main causes behind the decline of insect pollinators is the loss and fragmentation of habitats. Bee species diversity is related to both cover and diversity of flowering plants. Most of the forests in Sweden are actively managed for timber production and this has an effect on the forest understory and subsequent on the insect species that occur in the forest areas. Forest restoration aims to initiate, assist or accelerate the recovery of an ecosystem. It strives to restore the presumed historical composition, structure, function, productivity and species diversity of an ecosystem present at a stand. This study aims to examine the effect of prescribed burning and gap creation, two recently introduced forest restoration and management tools, on bumblebees, flowering plants and their interactions. In 20 boreal forest stands in northern Sweden two restoration treatments, prescribed burning and artificial gap creation, were assigned in 2011, untreated stands were used as references, 15 stands were chosen for this study; 5 stands of each treatment and 5 control stands. The bumblebees were trapped using pan and window traps and the vegetation was recorded in 12 of the 15 stands. A total of 130 bumblebee individuals were caught belonging to 7 species. *Bombus pratorum* was the most abundant and is the most commonly found species in Sweden. In the burned stands higher number of individuals and species were caught compared to the gap cutting treatment and the control stands. An increase in abundance can be explained by the increased temperatures in the burned stand enabling the bumblebees to be more active in these areas, while using less energy. There was no difference found between the treatments and the cover and the production of blueberry (*Vaccinium myrtillus*) and lingonberry (*Vaccinium vitis-idaea*). An explanation for this is that the burning and gap cutting treatment was performed in 2011 and that during the first 2 years after a fire disturbance there is a peak in productivity in the area, which over the following year's declines back to its original state. I show that prescribed burning can be beneficial for bumblebees, it increases their abundance and species richness compared to gap cutting and the control forests. The bumblebees were caught with two different trap types; coloured pan traps and coloured window traps; in the colours yellow, blue and white. The colour blue and yellow were the most effective colours, which could be explained by the types of flowers on the forest floor. The coloured window traps were the most effective trap type; this can be simply due to the added window; which enlarges the chance of bumblebees being caught. Window traps in the colour blue and white were the most effective method to catch bumblebees in this study. However it is best to use the colours blue, white and yellow to enhance the probability of catching different bee species. My study shows that the boreal forest is a utilized habitat by bumblebees and should thus be managed in a sustainable way. This study shows that prescribed burning is beneficial for bumblebees and this positive effect might also transcribe to other insect pollinators.*

Keywords: bumblebee, *Bombus* spp., forest restoration, abundance, species richness, species composition, boreal forest

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1 Introduction

Pollinators are an important part of biodiversity and pollinating insects, especially bees, are the primary pollinators of plants. Of all the wild plant species about 80% are reliant on insect pollinators (Potts et al., 2010). Yet the number of pollinating insects is declining all over the world (Potts et al., 2010, Goulson et al., 2002). Mainly as a result of loss and fragmentation of habitats, increasing pesticide application and fertilization (Potts et al., 2010, Altieri and Nicholls, 2003) and the changes in the floral resources (Potts et al., 2010, Goulson et al., 2002). Plant – pollinator interactions are sensitive to changes in both size and spatial arrangement of plant populations (Mustajarvi et al., 2001). Bee (*Apidae*) species richness declines when there is a higher level of isolation between suitable habitats (Jauker et al., 2009). And a decline in pollinator diversity and abundance can cause a decline in the pollination services for wild plants. Bee species diversity is related to both cover and diversity of flowering plants (Holzschuh et al., 2007, Weibull et al., 2003). Bumblebees (*Bombus spp.*) are an important pollinator of wild flowers, but can also pollinate pollinator-dependent crops (Vereecken, 2017, Michener, 2007).

Bees in the boreal forest are important pollinators of *Vaccinium* spp. and *Salix* spp. (Kevan and Baker, 1983). Forests cover 60% of the land area in Sweden (Lundmark et al., 2014) and most of the forests in Sweden are actively managed for timber production (Thomas et al., 1999). Over the last 100 years silvicultural methods have become more effective and as a result the forest standing stock has doubled (Lundmark et al., 2014) while structures important for biodiversity like old trees and coarse dead wood have declined (Niemela, 1997). To protect the biodiversity in Swedish forest, environmental policies state that the forests should be managed in a way that all the natural occurring species are maintained in viable populations (Angelstam et al., 2011). Half of the threatened species in Sweden are sensitive to thinning and clearcutting (Berg et al., 1995) and current logging practices negatively affect species that rely on old growth forests (Niemela, 1997).

Different forest management measures including logging affect the forest understory (Thomas et al., 1999) and subsequently the insect species that occur in the forest (Niemela, 1997). The pollinator species richness increases with the decrease of tree basal area and increased percentage of herbaceous cover (Campbell et al., 2007). Most of the flower-visiting insects in the boreal forest belong to the

orders *Hymenoptera* and *Diptera* (Nielsen, 2007). A lot of species of *Hymenoptera* favour open habitats with more sunlight and higher temperatures (Campbell et al., 2007). The same relationship is found in bees (*Apiformes*), the higher the forest cover the lower the bee abundance and species richness (Winfree et al., 2007). The number of bumblebees increases after logging because the amount of flowering plants increases, due to the increased availability of light. (Pengelly and Cartar, 2010, Romey et al., 2007).

Studies have shown that bee and wasp (*Vespidae*) species richness increases in clear-cuts (Rubene et al., 2015a), that insect species richness increases in thinned forest patches (Thomas et al., 1999), that beetle (*Coleoptera*) species richness increases in gap-cutting stands (Hägglund et al., 2016, Hjältén et al., 2017) and that there is a difference in insect species composition in burned and unburned stands (Rubene et al., 2015a, Hägglund et al., 2016, Hjältén et al., 2017, Mateos et al., 2011). This is a response to the differences in plant cover, species richness and microclimate. Flower rich and early successional stands in boreal forest landscapes are especially important for bee and wasp species (Rubene et al., 2015a).

In forest ecosystems bees have been associated with natural disturbances such as forest fires and river floodplain disturbances. These disturbances lead to the creation of early successional habitats that are used by bee species. Anthropogenic disturbances have replaced these natural disturbances (Winfree et al., 2007). Forest restoration aims to initiate, assist or accelerate the recovery of an ecosystem. It strives to restore the presumed historical composition, structure, function, productivity and species diversity of an ecosystem present in an area (Ciccarese et al., 2012). Before the forests became heavily managed, wildfires used to be a normal occurrence in the Swedish forests. However after the forest became actively managed for timber production fire prevention efforts went up and subsequently the fire frequency went down. Nowadays burning is used as a tool for forest restoration (Nilsson, 2005) and prescribed burning has a positive effect on fire dependent invertebrates (Niemela, 1997).

A common change depending on the already existing dwarf-shrubs layer is that after a fire disturbance this layer changes from a high cover of crowberry (*Empetrum nigrum*) to a high cover of lingonberry (*Vaccinium vitis-idaea*) and blueberry (*Vaccinium myrtillus*) (Nilsson and Wardle, 2005). Previous studies show responses of pollinators to forest fire and the regeneration of flowers is closely followed by the reoccurrence of bee species. (Potts et al., 2003). In a Spanish forests reserve, which is dominated by holm oak trees (*Quercus ilex*) and pine trees (*Pinus halepensis* and *Pinus niger*) with a holm oak understory vegetation, a fire disturbance increased the number of Hymenoptera families found in the area (Mateos et al., 2011). In an Aleppo pine forest (*Pinus halepensis*) in Israel which was in various stages of post-fire regeneration it was observed that after a fire disturbance there was immediately a short period in which there is a drop in the richness of flowering plants and bee species. The effect of the forest fire in the Mediterranean on the vegetation and insect abundance stabilises after the first couple of years and afterwards returns to

its previous state during the following 50 years (Potts et al., 2003). In a Mediterranean pine forest, the effect of fire was measured after 4 years. Here the forest fire strongly affected the community structure of bees, beetles, and sawflies (*Symphyla*) (Lazarina et al., 2017).

In the boreal forest gap disturbance contributes to the structure, function and species diversity (Kuuluvainen, 1994) and as a restoration measure, gap cutting can be used to simulate small scale gap dynamics to increase structural variation and dead wood volume in a stand (Hägglund et al., 2015). These measures have in common that they create a less dense forest canopy and might have similar effects on flower visiting insects. Thinning of the trees in Douglas-fir (*Pseudotsuga menziesii*) plantations in Washington State (USA) has a positive effect on the vegetation in the understory. With a higher level of thinning the cover of vegetation in the understory also increases. Insect species richness followed with a similar response to thinning (Thomas et al., 1999). A thinning experiment in Japanese cedar (*Cryptomeria japonica*) plantations concluded that thinning affected insect community structure. Insect species richness and abundance increased 1 year after thinning. After 3 years, there was only a difference in species richness of bees, the abundance of bees, butterflies (*Lepidoptera*), and hoverflies (*Syrphidae*) compared to the control (Taki et al., 2010).

Most of the insects used in forest biodiversity studies focus on beetles and not a lot is known about other taxa (Rubene et al., 2015b) and few studies have been performed on the importance of bumblebees in forest ecosystems. This pilot study is one of the first studies that examines bumblebee assemblages in boreal forests. The aim of this study is to examine the effect of burning and gap creation on the occurring bumblebee species and how the flowering plant species composition affects the bumblebee species. The two main research questions asked are; is there an effect of burning and gap creation on the bumblebee species richness, abundance and composition? And, is there an interaction between of burning and gap creation and flowering plant species composition and bumblebee species richness? In line with previous research I predict that there will be more individuals present in the gap cut and burned stands compared to the control stands. And that there will be a difference in species composition, number and/or evenness between the control and the gap cut and burned stands. I predict that in the burned stands there will be a higher cover and productivity of the dwarfs-shrub layer.

Because there is not a lot of bee and bumblebee research performed in the boreal forest, I will use two different trapping methods and three different colours to assess which method is the most effective in this type of environment. The methods commonly used for trapping insect pollinators are pan traps, netting, observation plots, trap nests and flight interception traps of various kinds. Pan traps are bowls, insects approach the bowls, land on the water, and drown (Roulston et al., 2007, Cane et al., 2000). Window traps consist of a bowl and glass, insects will fly against the glass, fall into the tray and drown (Kearns and Iouye, 1993). The catchment trays can be used in several colours (for example blue, yellow, white and red) and the

colours white, yellow and blue have been proven to be effective to trap different species of bees (Toler et al., 2005). I will test the effectiveness of pan traps and coloured window traps and the effectiveness of the colours yellow, blue and white. The aim is to find the most effective combination of trap and colours. And the two sub questions are; which trapping method is more effective in a forest setting? And, which colour is more effective in a forest setting? I predict that window traps and the colour yellow will be most effective.

2 Materials and methods

2.1 Bumblebees

Bees are the most adapted *Insecta* to anthophily, they are highly diverse behaviourally and taxonomically. Their mouthparts are highly adapted for feeding on nectar and their bodies for collecting pollen, bees are almost completely vegetarian (Kevan and Baker, 1983). There are approximate 300 bee species in Sweden (Nilsson, 2007), including 40 species of bumblebees (*Bombus spp.*) (Mossberg and Cederberg, 2012), the honey bee (*Apis mellifera*) and more than 250 solitary bee species. New species, estimated to be 1-2 per year, move into Sweden from the south (Denmark) (Nilsson, 2007, Løken, 1973), and/or from the east (Finland) (Løken, 1973). Bumblebees occur in temperate climates around the world, with only a few exceptions (Matthews and Matthews, 2010). Bumblebees can be active longer during the season compared to other insects because they are partial endothermic i.e. able to generate their own heat (Willmer and Stone, 2004). Social bumblebee colonies are founded by a single overwintering queen. The queen stocks pollen and honey separately from the brood cells in special wax storage pots. She then lays several eggs in a single wax brood cell, the first brood will result in workers that enable the queen to stay inside the nest and lay more eggs. Later in the season the queen stops producing workers and starts producing males and new queens. Once mated the new queens find a good place to hibernate until the next spring (Matthews and Matthews, 2010).

2.2 Experimental stands

During 2011 two restoration treatments were assigned to 20 forest stands each. An additional ten forest stands were chosen as untreated references. The two treatments were restoration burning and artificial gap creation. Artificial gap creation was used to simulate natural gap dynamics in the forest stands. (Hägglund et al., 2015, Hägglund and Hjältén, 2016, Hägglund et al., 2016, Hjältén et al., 2017).

Out of these stands 15 forest stands were chosen for this study, 5 stands of each treatment and 5 control stands. The stands were clustered in groups of 3 so that the stands with different treatments were close to each other this was possible for 4 out of the 5 groups. The stands (figure 1) varied from 3.6 to 21 ha and were dominated by Scots pine (*Pinus sylvestris*) and/or Norway spruce (*Picea abies*). Downy birch (*Betula pubescens*), silver birch (*B. pendula*), aspen (*Populus tremula*) and goat willow (*Salix caprea*) occurred scattered throughout the stands. The dominant forest

type in all stands was mesic dwarf-shrub with blueberry (*Vaccinium myrtillus*) as the dominant species in the field layer (Hägglund et al., 2016, Hjältén et al., 2017).

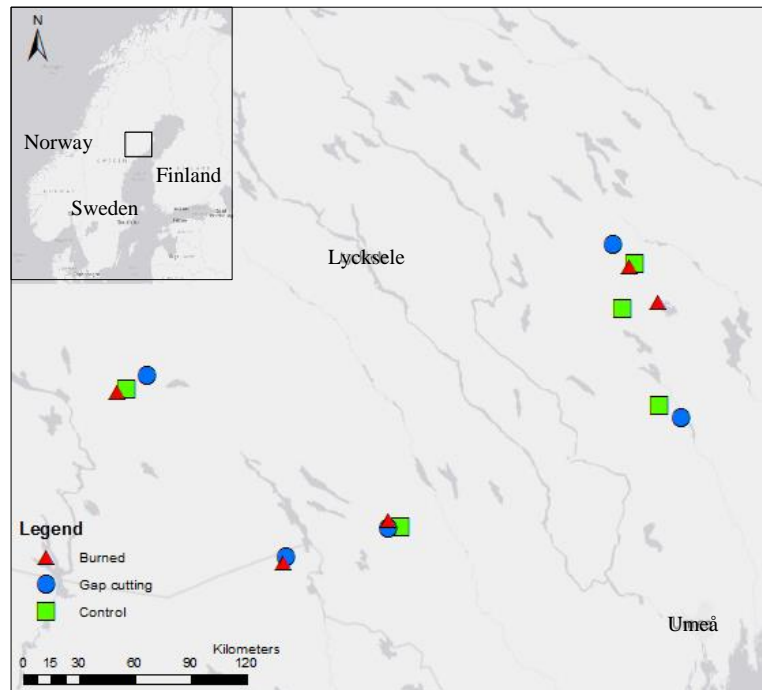


Figure 1 Map of the locations selected for the study with the 3 different treatments

2.3 Method

For this study I trapped bees with two different trapping methods and I did a vegetation inventory which included the netting of passing bumblebees. The data was collected over a period of 4 weeks, I made sure that the blueberries had started flowering and the traps were put out in the second week of June. Generally June is the month when bumblebees are most active and the colonies are at its largest (Söderström, 2013). When the traps were emptied the samples were put in separate containers for each trap colour. The caught insects were stored in 70% alcohol. The bees were separated and washed, dried and pinned for identification.

2.3.1 Vegetation inventory

The understory in the northern Swedish boreal forests consists mainly of ericaceous dwarf shrubs, feather mosses and reindeer lichens. The three most common and abundant dwarf-shrub species are blueberry (*Vaccinium myrtillus*), lingonberry (*Vaccinium vitis-idaea*), and black crowberry (*Empetrum hermaphroditum*) (Nilsson and Wardle, 2005). The vegetation inventory was performed together with the bee plots. The assumption was that the floral availability would resemble the recorded plot. The vegetation was sampled in each stand around the traps using 3 sample plots with a size of 1x1 meter. The following was recorded;

- Date, time, weather (sunny, partly cloudy, cloudy), temperature,
- General vegetation structure, 0 = Open – 4 = closed,
- Flower/plant species,

- Number of flowers,
- Any other flowering plant species within a 20 meter radius of the plot.

2.3.1 Insect inventory

There are different methods developed to sample bee species, most commonly used are pan traps, netting, observation plots, trap nests and flight interception traps of various kinds. Pan trapping and netting are recommended as a solid method for catching bees by several other studies (Campbell and Hanula, 2007, Cane et al., 2000, Nielsen et al., 2011, Popic et al., 2013, Rubene et al., 2015b, Toler et al., 2005, Westphal et al., 2008, Wilson et al., 2008). Pan trapping has several advantages compared to netting, it is simple and therefore there is no need for skilled net collectors. The method has proven to be particularly good at catching numerous species of bees (Cane et al., 2000, Roulston et al., 2007, Westphal et al., 2008).

Two different trapping methods were used to collect the samples. Coloured pan traps and coloured window traps. In this study the traps were filled with a mixture of water, dish soap and propylene glycol (to preserve the insects). The traps were filled with the mixture for 1/3 to prevent the traps from overflowing when it rains. To further prevent the traps from overflowing drainage holes were punctured in the trays. The traps were emptied after two weeks and were set out for a period of 4 weeks. In the each of the stands 3 window traps and 3 pan traps are set up, resulting in 6 traps with 15 trays in total in each stand. The location of the traps was randomly chosen from the centre of the stand and placed at least 20 m apart. Bee plots were conducted to assess whether the traps were effective in catching the species present in the stands.



Figure 2 Example of a pan trap on the left and a window trap on the right (Photo: Raisja Spijker)

Pan traps – Pan traps are bowls which are filled with soapy water (Roulston et al., 2007, Cane et al., 2000), or with water and propylene glycol (to preserve the insects) (Rubene et al., 2015a), insects approach the bowls, land on the water, and drown (Roulston et al., 2007, Cane et al., 2000). Pan traps can be used in many colours (for example blue, yellow, white and red) and the colours white, yellow and blue have been proven to be effective to trap different species of bees (Toler et al., 2005). In this study the traps consisted of aluminium trays ($\pm 13 \times 10$ cm) with on the inside three different colours; blue, yellow and white (Biltema hobbyfärg (spray paint) art.no.: 36418, 36516 and 36565) mounted to a plastic pole, figure 2, the pan traps were set up with the tray at vegetation height. The traps were filled with a mixture of water, dish soap and propylene glycol.

Window traps – The catchment trays of a window trap work the same way as a pan trap. The insects will fly against the glass, fall into the tray and drown (Kearns and Iouye, 1993). In this study the traps consisted of a Plexiglas plate (10 x 20 cm) with an aluminium tray ($\pm 13 \times 10$ cm) attached beneath. These aluminium trays were also spray painted with one of the three colours mentioned above. The trays were paired and the traps consisted of the following three colour combinations; white/blue, white/yellow and blue/yellow, figure 2, the window traps were also set up with the tray at vegetation height. The traps were filled with a mixture of water, dish soap and propylene glycol.

Bee plots – The bee plots were conducted after and on the same spot as the vegetation inventory so that the data recorded for the vegetation inventory could also be used for the bee plots. The bee plots were a deviation from the standard bee transects as described by Prÿs – Jones and Corbet (2011) but it resembled the bee walks from Carvell (2002). The bee observations were done once in each stand, directly after the vegetation was sampled between 9.00 and 19.00. All species of bumblebees visiting or passing within a 20 meter radius of the vegetation sampling plots were caught with a net, for 20 min. All bumblebees were collected and brought to the lab for determination. For all bumblebees, the following was recorded; the plant species it was found on or if bee caught in flight.

The bumblebees were determined to species level. To preserve the bumblebees, they were pinned (mounted on insect pins), labelled and stored in insect boxes. Three labels were used, the first label included species name and name person who identified the insect. The second label included the date, stand of collection, name of collector. And the third label included additional data such as the plant on which the insect was collected (Kearns and Iouye, 1993).

2.4 Data analysis

The bumblebees caught with the netting method during the bee plots were not included in the statistical analysis because of the difference in effort and low numbers caught compared to the trapping method. Furthermore this method is prone to several biases such as temperature, time of the day, person netting etc. The bumblebees caught with the netting method were used to compare to the species to the species caught with the traps to investigate if there was no big difference in species caught using either method. Because of limitations in time and bad weather conditions 12 out of the 15 plots were visited for the vegetation inventory and bee plots. The vegetation data from the vegetation inventory was put together as an average for each stand and used for further analysis. For the statistical analysis the program R studio and package VEGAN was used.

The species diversity index (D) was calculated with the formula for the Simpsons Diversity Index. Where n stands for total number of individuals of a species and N for total number of individuals of all species. The outcome of D was converted into the Simpson's Reciprocal Index (1/D) resulting in the higher the value the greater the diversity in the community is.

$$D = \sum \frac{(n - 1)}{N(N - 1)}$$

To examine whether there was a difference in species composition between the stands and treatments the Sørensen's Coefficient (CC) was used to calculate the similarity between the stands. Where C stands for the shared species, S1 for the no. of species in stand 1 and S2 for the no. of species in stand 2. The closer the value is to 1, the more dissimilar the species composition is.

$$CC = \frac{S1 + S2 - 2C}{S1 + S2}$$

To calculate the difference in species composition between the treatments an ANOSIM was used. The ANOSIM used the values calculated by the Sørensen's Coefficient and pair wise testing using ANISOM was conducted to further explore the significant difference.

The difference between the dependent variables and the different treatments was analysed with an ANOVA in combination with Tukey's post hoc test, this method was used for both vegetation and bee richness data. The assumption of normality and homoscedasticity were checked with the use of Shapiro-Wilk test and the Bartlett test, if the assumption was not met a Kruskal-Wallis test was performed instead of an ANOVA. A linear regression was performed to examine the effect of the vegetation variables on the bumblebee individuals, species and individuals. The

assumptions about the normality of the residuals was checked with the Shapiro-Wilk normality test.

To test whether the effectiveness of the colours is dependent on the trap kind a chi-squared test was performed. The difference in number of bumblebees caught between the 3 trap colours was tested with a binominal exact test because the number of traps per colour were equal. And to test whether there was a difference in the effectiveness of the 2 different kinds of traps a binominal test was used to compare the two proportions in each trap, because the number of traps in each category was not equal.

Table 1 Overview of bumblebee species caught per method per treatment

Species	Trapping			Netting		
	Burning	Control	Gap	Burning	Control	Gap
<i>Bombus bohemicus</i>	3	2	2	1	1	2
<i>Bombus cingulatus</i>	1	1	1	-	-	1
<i>Bombus hortorum</i>	7	3	2	4	1	3
<i>Bombus hypnorum</i>	5	2	4	1	1	-
<i>Bombus lucorum coll.</i>	8	1	-	2	2	3
<i>Bombus pascuorum</i>	4	1	5	1	-	3
<i>Bombus pratorum</i>	34	20	24	5	3	3
<i>Bombus soroeensis</i>	-	-	-	1	-	-
Total	62	30	38	15	8	15
Grand Total	130			38		

3 Results

3.1 Overview of the caught bumblebees

In the traps a total of 130 bumblebees were trapped, which belonged to 7 species. During the netting a total of 38 bumblebees were caught, belonging to 8 species, the number of individuals caught of each species is visible in table 1Table , *Bombus soroeensis* was the only species that was only caught using the netting method. In figure 3 there is an overview of the species caught in the traps in each stand divided by treatment. Most noticeable is that it that *Bombus pratorum* was the most abundant species and that *Bombus lucorum coll.* was found in all the burned stands, one control stand and not in the gap cutting stands.

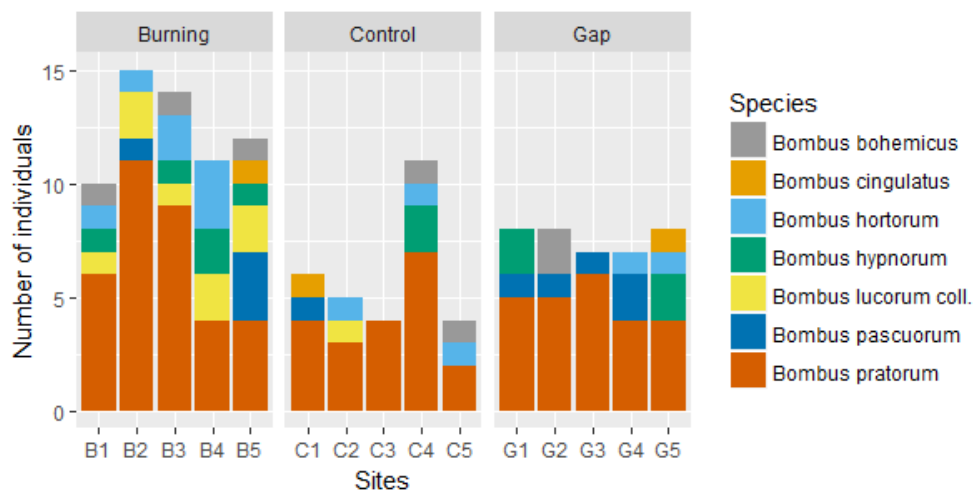


Figure 3 Bumblebee species composition of the specimens caught in the traps

3.2 Effect of forest restoration on bumblebees

The Venn diagram in figure 4 shows that there was no big difference in the total number of species trapped between the treatments, there was not a single species that was unique to one of the three treatments. There was a significant difference in species composition between the treatments ($R=0.230$, $p=0.049$, ANOSIM). The pair wise testing showed that there was a significant difference between the burned and gap cutting treatment ($R=0.556$, $p=0.018$, ANOSIM) and not between the burned and control treatment ($R=0.140$, $p=0.100$, ANOSIM) and control and gap cutting treatment ($R=0.010$, $p=0.446$, ANOSIM).

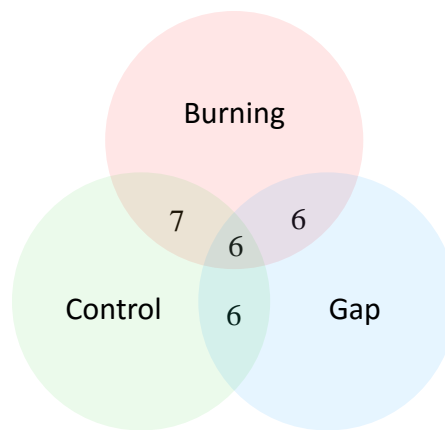


Figure 4 A Venn diagram showing the overlap in species between the treatments. The diagram shows the overlap of the species between the treatments.

The number of bumblebees trapped in the traps differed significantly between the different treatments ($\chi^2_{(2)}=9.311$, $p=0.009$, figure 5A). There was a significant higher amount of bumblebees trapped in the burned stands than in the control stands ($Z=2.989$, $p=0.008$) but the number of caught bumblebees was not significantly different between the burned and gap cutting treatment ($Z=2.028$, $p=0.06$) and the gap cutting treatment and control stands ($Z=-0.961$, $p=0.338$).

The number of bumblebee species trapped in the traps was also significantly different between the treatments ($F_{(2)}=7.583$, $p=0.007$, figure 5B). Again there was significantly higher amount of species trapped in burned stands than in the gap cutting stands and control stands ($p=0.020$ & $p=0.011$) and no difference between the gap cutting stands and the control stands ($p=0.934$). There was no significant difference in species diversity in between the two treatments and the control stands ($F_{(2)}=0.566$, $p=0.582$, figure 5C).

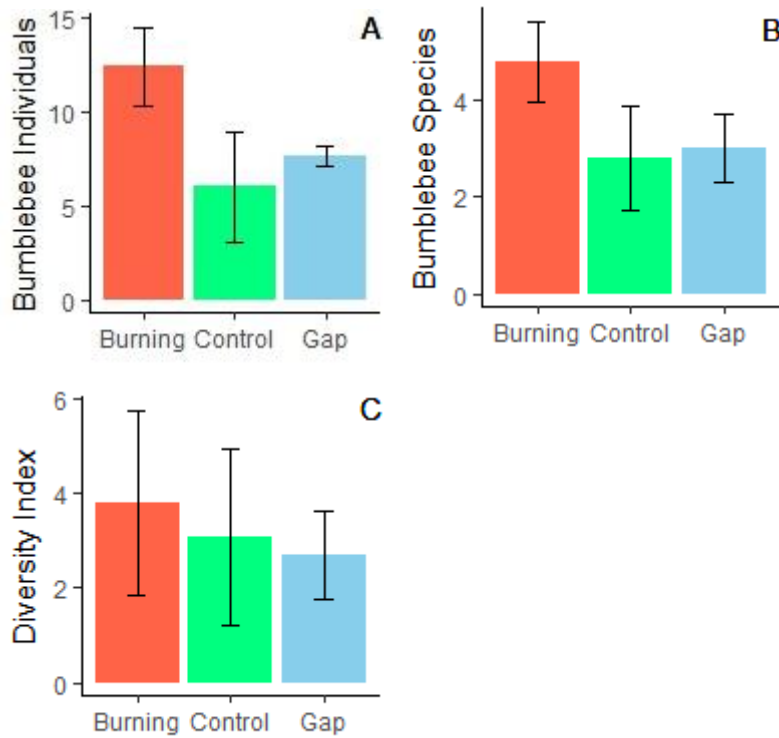


Figure 5 Effect of the treatments on the bumblebees. A shows the effect of treatment on the number of individuals caught, B shows the effect of the treatment on the number of species caught and C shows the effect of treatment on the diversity.

3.3 Effect of forest restoration on blueberry and lingonberry

There was no significant difference found between the blueberry (*Vaccinium myrtillus*) cover ($F_{(2)}=0.699$, $p=0.201$, figure 6A), the number of blueberry flowers ($F_{(2)}=0.711$, $p=0.517$, figure 6C) and the lingonberry (*Vaccinium vitis-idaea*) cover ($F_{(2)}=1.535$, $p=0.267$, figure 6B) in relation to the two treatments and the control stands.

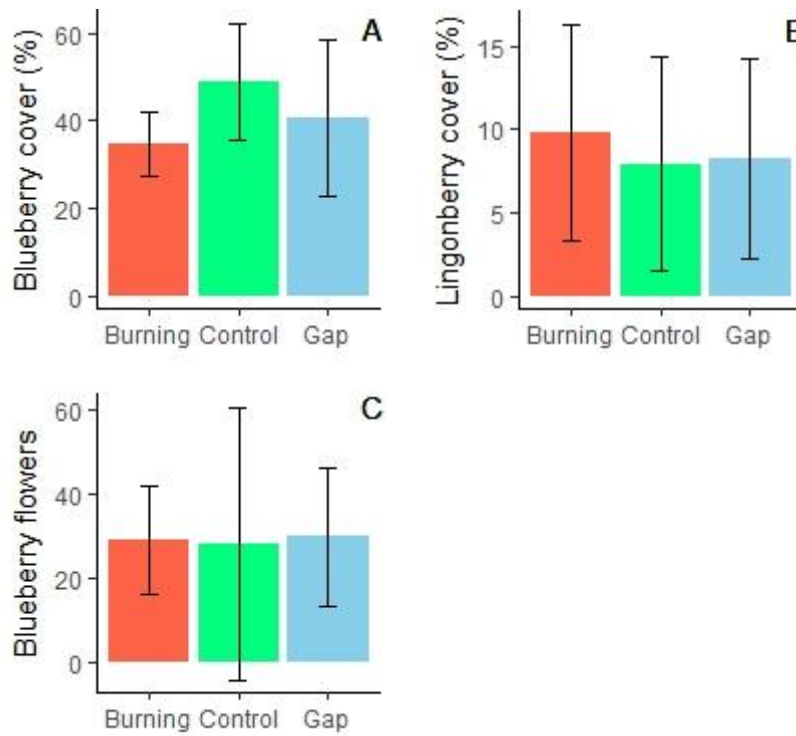


Figure 6 Effect of treatment on blueberry and lingonberry. A shows the effect of treatment on the percentage of blueberry cover, B shows the effect of treatment on the percentage of lingonberry cover and C shows the effect of treatment on the number of blueberry flowers.

3.4 Effect of the understory vegetation on bumblebees

There was no linear relationship between the number of bumblebee individuals trapped and the percentage of cover of blueberry bushes ($F_{(10)}=0.754$, $p=0.406$) and number of blueberry flowers ($F_{(10)}=0.842$, $p=0.380$). There was also no relationship found between the number of bumblebee species trapped and the percentage of cover of blueberry bushes ($F_{(10)}=1.818$, $p=0.207$) and the number of blueberry flowers ($F_{(10)}=0.667$, $p=0.433$).

3.5 Effectiveness of the traps

In table 2 there is an overview of the number of bumblebees trapped in the trap per trap model and colour. Trap colour and trap kind were independent from each other ($\chi^2_{(2)}=1.8328$, $p=0.4$, chi-squared test). Thus the effectiveness of trap type and trap colour could be tested separately. There was a significant difference in the number of bumblebees trapped between the colour yellow and blue ($p<0.001$, binomial exact test) and the yellow and white ($p<0.001$, binomial exact test), with the colour yellow trapping the lowest amount of bumblebees. There was no significant difference in the amount of bumblebees trapped between the colour blue and white ($p\text{-value} = 0.771$, binomial exact test). There was a significant difference between the number of bumblebees trapped between the two traps types ($\chi^2_{(1)}=38.43$, $p<0.0001$, binominal test).

Table 2 Overview of the number of bumblebees caught in the different trap types per colour

	Blue	Yellow	White	Row totals
Pan trap	25	12	18	55
Window trap	30	12	33	75
Column totals	55	24	51	130

4 Discussion

4.1 Bumblebees in the forest

The forest might not be the preferred habitat for some bumblebee species, while others use the forest to forage for nectar and pollen (Kreyer et al., 2004). There are plants in the forest that rely on the pollination of bumblebees (Kevan and Baker, 1983, Vereecken, 2017, Michener, 2007). Since 60% of Sweden is covered with forest (Lundmark et al., 2014), it is an important habitat to consider for biodiversity conservation. Certain tree species and flowers in the forest understory are used by bumblebees when there is no or too little other plants flowering (Ulyshen et al., 2010) and four of the seven bumblebee species found during this study (*Bombus pratorum*, *Bombus cingulatus*, *Bombus lucorum coll.* and *Bombus bohemicus*) are important pollinators of *Salix spp.* and *Vaccinium spp.* (Mossberg and Cederberg, 2012). So although the bumblebee densities in forests are low compared to heathland, it is a habitat that they utilize and it is probably important due to the large area covered. With the current forest management practices stands get denser and are even-aged (Lundmark et al., 2014). This means that forest restoration is important for the conservation of bumblebees and other insect species. From the results of this study it can be suggested that the species *Bombus cingulatus*, *Bombus lucorum coll.* and *Bombus hortorum* benefit from a more open forest either due to prescribed burning or gap cutting.

4.2 Effect of forest restoration on bumblebees: treatment

The abundance of individuals and species richness differed between stand types. In the burned treatment there was a higher number of individuals and species caught compared to the gap cutting treatment and the control and the gap cutting treatment was similar to the control. By contrast, there was no difference in the species diversity (Simpson's Diversity Index) between the treatments and control stands. One explanation for this could be that the Simpson's Diversity Index gives more weight to the more abundant species in a sample and *Bombus pratorum* is a very abundant species in all the stands, which is visible in figure 3. In the gap-cutting treatment stands only 6 species were found in contrast to 7 species in both the control as in the burned treatment stands. A difference in species composition was found between the burned and the gap cutting treatment. This means that the gap-cutting stands had less species in common with the burned stands than the control stands.

The differences between the stands could be affected by differences in the understory vegetation as a result of the restoration treatments (Potts et al., 2003, Nilsson and Wardle, 2005) but also by the higher temperature in the burned stands. In a study on bird response to ecological restoration (Boer, 2017), temperature loggers were placed in and outside nest boxes to measure the temperature in some of the same stands used by this study. In the burned stands the temperature inside and outside the nest boxes was significantly higher than in gap and control stands and there was no difference in temperature between gap and control stands (Boer, 2017). Because bumblebees are cold blooded, as a result their metabolism and activity is influenced by the temperature of their bodies, which temperature is almost entirely dependent on that of the surrounding environment. So a low temperature inhibits activity, whilst a higher temperature stimulates activity (Mellanby, 1939). So more bumblebee individuals and species found in the burned stands than in gap and control stands and the fact that there is no difference between the gap and control stands might be explained by increased temperature in the burned stands enabling the bumblebees to be more active in these areas, while using less energy. While there being no temperature advantage in the gap cutting and control stand, as I observed there to be not a lot of difference in canopy cover between the two.

4.3 Bumblebee species

All of the 7 bumblebee species (*Bombus spp.*) found during this study are commonly found in forests, and a few are important pollinators of blueberry (*Vaccinium myrtillus*) and *Salix spp.*

During this study *Bombus pratorum* was found throughout all the stands, however in the burned stands there was a slightly higher number of individuals. Suggesting a positive effect of the burned treatment on this species. It is the most abundant species in all the stands compared to the other species. *B. pratorum* is wide spread all over Sweden, it is found in all types of forests. But can also be found in meadows, pastures and forest edges. It is an important pollinator of blueberry and lingonberry (*Vaccinium vitis-idaea*) and raspberry (*Rubus idaeus*) (Mossberg and Cederberg, 2012, Söderström, 2013). Their high numbers caught during this study makes clear that they do forage on the blueberry and later on lingonberry in the boreal forest.

Of the species *Bombus cingulatus* three individuals were trapped, two in gap cutting stands and one in a control stand. *B. cingulatus* is a bumblebee found in middle and northern Sweden found and is a specialist in the coniferous boreal forest. It prefers a more open forest habitat and the queen mainly feeds on blueberry (Mossberg and Cederberg, 2012, Söderström, 2013).

The species *Bombus hypnorum* was trapped evenly all over the different treatments, this is in line with the species being a generalist. *B. hypnorum* is an all-round species found all over Sweden that is found near human activity but also in the forests in the mountains (Mossberg and Cederberg, 2012, Söderström, 2013).

Bombus lucorum coll. was mainly trapped in the burned treatments and only in one control stand. *B. lucorum coll.* is an all-round species found all over Sweden. When they first emerge, they feed on *Salix* spp. and later on mainly on blueberry (Mossberg and Cederberg, 2012, Söderström, 2013). So even though this species is supposed to be a generalist, and should thus thrive in different environments, prescribed burning seems to have a positive effect on the occurrence of this species.

Bombus pascuorum was found all over the different treatments, which is in line with the species being a generalist. *B. pascuorum* is found all over Sweden in all environments except from pronounced farmland and forages on a broad range of species (Mossberg and Cederberg, 2012, Söderström, 2013).

Most individuals of *Bombus hortorum* were trapped in the burned stands, suggesting a positive effect of the burning treatment on this species. *B. hortorum* is found all over Sweden in broadleaved and mixed forests, but also near human activity (Mossberg and Cederberg, 2012, Söderström, 2013). The species seems to benefit from the burned treatments making the boreal forest more habitable for this species.

Individuals of *Bombus bohemicus* were evenly spread throughout the treatments. *B. bohemicus* is the most common cuckoo bumblebee in Sweden and is found in a wide range of habitats. A cuckoo bumblebee female doesn't collect her own pollen and nectar and doesn't produce any workers. She takes over a social bumblebee colony and here she feeds on the stored pollen and nectar but also on the eggs and larvae from the queen. *B. bohemicus* parasites mainly on *B. lucorum coll.* and prefers to feed on blueberry and lingonberry nectar (Söderström, 2013). Since the same amount of individuals was found throughout the treatments it suggests that *B. lucorum coll.* should also be present in these stands or that they choose to parasite on other species as well. They are believed to also parasite on other species that have a similar colour pattern as the queen does (Söderström, 2013). Their presence throughout all the treatments could be explained by the presence of *Bombus hortorum*, which has a similar color pattern as *B. lucorum coll.*, *B. hortorum* was found throughout all the treatments as well.

4.4 Effect of forest restoration on bumblebees: vegetation

After a fire disturbance the amount of flowers increases and the abundance of bees follows this trend (Potts et al., 2003). However, I found no difference in the vegetation between the different treatments. One explanation could be that the restoration treatments were done in 2011 and the vegetation in the stands were no longer experiencing the effect of the fire. I did observe the burned stands to be more open and warmer and the blueberry bushes were smaller compared to the gap cutting and control stands. However no difference was found in the overall cover of blue- and lingonberry or number of flowers between the different treatments. (Potts et al., 2003) shows that in forests in the Mediterranean during the first 2 years after a fire disturbance there is a peak in productivity and insect diversity in the area, which over the following 50 years declines back to its original state. In the boreal

forest the net primary production (NPP) recovers after about 9 years depending on the intensity of the fire (Hicke et al., 2003). That the restorations have been performed in 2011 could explain the lack of fire effect on the vegetation variables.

There was also no significant relation found between the vegetation variables and number of individuals/species of bumble bees. However (Brosi et al., 2008) also found no effects of flowering plant species and number of flowers on bee abundance. This could be explained by the fact that bumblebees benefit the most from the fire the first couple of years when the areas are flower rich and in their early successional state (Rubene et al., 2015a). The method used to sample the vegetation during this study is not the usual way of surveying vegetation. The usual method for vegetation studies are several line transect in an area (Buckland et al., 2007), for this study there was limited time hence transects were not possible and 3 sample plots in each stand were taken instead. Which is might not have been enough to cover the diversity of the stand and as a result doesn't give a representable presentation of the understory in the stand. I observed that there was a difference in the size of the blueberry bushes but not the cover, so for future studies I recommend to also do an estimate of the field layer biomass.

4.5 The trap effect

With the trapping method 7 species were trapped and with the netting method 8 species were caught. The same species were caught using both methods and the 8th species caught with the netting method was, *Bombus soroensis* which is in general a rare species in Sweden (Bommarco et al., 2012). So overall the traps are representative of the species that occur on the forest floor.

The colour yellow was the least effective colour in catching bumblebees and the colours white and blue were equally effective. In the studies of (Geroff et al., 2014), on a Illinois prairie, and (Cane et al., 2000), in a Arizona desert shrub community, blue pan traps also outperformed yellow pan traps. However in a north eastern US forest; blue was the least effective colour (Giles and Ascher, 2006). In a north western Iowa grassland; yellow was the colour that was the most effective (Kwaiser and Hendrix, 2008) and in a Louisiana long leave pine savannah fewer bees were collected in white traps compared to blue and yellow traps (Bartholomew and Prowell, 2005). So the effectiveness of the colours vary greatly and may be dependent on both spatial and temporal factors (Geroff et al., 2014). Environmental factors, such as the colour of the blooming flora, could alter the effectiveness of the traps, the most abundant colour outcompete the trap colour (Hall, 2016). In addition some bee species have shown colour preferences (Toler et al., 2005). Hence it is common practice to use blue, yellow and white traps to enhance the probability of catching different bee species (Geroff et al., 2014, Vrdoljak and Samways, 2012). Pan traps are commonly used to survey pollinating insects as it is a more time efficient method than netting and doesn't require skilled field personnel (Cane et al., 2000). The window traps were not coloured in the study of (Rubene et al., 2015b) and were thus more effective for wood nesters. In

this study I added colour to the window traps which made them more effective for catching pollen collectors and it made them even more effective than pan traps.

The traps were placed at vegetation height, which is appropriate for sampling species that utilize the stands for foraging. However if the aim of the study is to sample all species occurring in the stands it is important to take into account that different bumblebee species have different flight and foraging patterns. Some might choose to fly over the forest canopy while others choose to use the flowers available on the forest floor (Mossberg and Cederberg, 2012, Kreyer et al., 2004) and placing only traps on the ground might not be sufficient to sample all the species occurring in the stand. In the study of (Tuell and Isaacs, 2009) they found that the number of bees captured in pan traps on the ground was lower than the number of bees trapped in the canopy. Even when they included lower stands with a blueberry cover the number of bees trapped on the ground did not change. This is also supported in the study of (Geroff et al., 2014) where elevated pan traps decreased the amount of traps needed to be able to estimate the bee species richness. Also in the study of (Ulyshen et al., 2010) the bees were more abundant in the canopy than on the ground. Bees frequent the canopy, especially during the time there is low nectar and pollen availability (Ulyshen et al., 2010). During this study the blueberry bushes were at their peak production so it was expected that most of the bumblebees would spend more time close to the forest floor.

The traps can be improved by enlarging the catchment trays since the catchment trays used in this study are relatively small and (Wilson et al., 2016) shows that bigger traps catch more specimens. The use of elevated pan traps can also increase the sample size (Ulyshen et al., 2010, Geroff et al., 2014, Tuell and Isaacs, 2009). To further increase the sample size it is possible to sample the whole bumblebee season. When using pan traps this does not increase the sample effort by a lot, since when the pan traps are of good quality and of larger size they could stay out for the whole season and be collected at the end. This will increase the probability of catching all the species present in the given area. Netting is not a preferable method in a forest habitat, netting is time consuming on its own and in the forests there is a low abundance of bumblebees present which makes the netting method even more time consuming. I recommend the following design for sampling the bumblebees in the boreal forest; the use of window traps with large coloured trays in the colour blue and white, covering a large area within the stand since the abundance of bumblebees is low, to use at least 10 pairs of traps (preferably more) and to start sampling at the beginning of the flowering period to the end of the bumblebee season. Yellow catchment trays can be added to enhance the probability of catching different bee species.

4.6 Summary

This study shows that prescribed burning is beneficial for bumblebees in the boreal forest, it increases their abundance and species richness compared to gap cutting and the control forests. That could be because flower rich and early successional sites in boreal forest landscapes are important for bee and wasp species (Rubene et al., 2015a). Prescribed burning also opens up the forest canopy more, increasing the amount of sunlight that can travel to the understory and in its turn increasing the temperature. Bumblebees and other insects benefit from a higher temperature and can be more active without using more energy. Window traps in the colour blue and white were the most effective method to catch bumblebees. However it is best to use the colours blue, white and yellow to enhance the probability of catching different bee species.

4.7 Management implications

Burning is the preferred method of restoration because it increases the abundance of bumblebees more than artificial gap creation does. Prescribed burning and gap-cutting opens up the canopy, increasing light and subsequent the temperature, allowing bumblebees to be more active while using less energy. During the restoration treatments in 2011 the artificial gap creation might not have been sufficient in creating large enough gaps in the canopy to differ from the control stands. Early successional stands in the boreal forest are important habitats for bees especially if they have high flower species richness. The succession of burned stands in the boreal forest can change from higher in crowberry (*Empetrum nigrum*) cover to higher in blueberry (*Vaccinium myrtillus*) cover. Blueberry and lingonberry (*Vaccinium vitis-idaea*) are the plant species that are important for bumblebees in the forest. Burned areas restore themselves to their pre-burning state so in order to keep the benefits there must be regular disturbances (not necessarily prescribed burning), either natural or artificial, to keep the area in an early successional state. But even if the burned stands no longer have the light and temperature benefits and are no longer in an early successional state they can still be beneficial for the bumblebees due to the change in understory vegetation.

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